OK TO ENTER: /CMW/
11/01/2010

Attorney Docket No. 33581-US-PCT
LNG Docket No. 64617.US / C-6710.0.Germany

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant(s):	Viktor Menart et al.
Application No.:	10/583,157
Filing Date:	June 16, 2006
Confirmation No.:	5099
Title:	A Pharmaceutical Composition Comprising an Active Principal and Sulphobetaine
Examiner:	Woodward, Cherie Michelle
Group Art Unit:	1647

DECLARATION OF VLADKA GABERC-POREKAR UNDER RULE 132

I, Vladka Gaberc-Porekar, hereby declare and state based upon personal knowledge that:

1. I am a research scientist the Slovenian National Institute of Chemistry (NIC), in Ljubljana, Slovenia. In this position, I have frequently collaborated with employees of LEK Pharmaceuticals, D.D., the assignee of the present application. In particular, I worked on the development of the technology disclosed in the above-referenced patent application (together with Viktor Menart and Barbara Podobnik) under a mutual agreement between LEK and NIC.

2. In the current Office Action in this case, I understand that the Examiner has rejected Claims 1, 2, 10, and 12 as allegedly being anticipated by U.S. Patent Number 5,500,416 to Miyazawa et al. ("Miyazawa"). I respectfully disagree with the Examiner's contention that the claims are anticipated by the Miyazawa reference.

3. Claims 1, 2, 10, and 12, are directed to a pharmaceutical composition for parenteral administration which comprises an active pharmaceutical ingredient and a non-detergent sulfobetaine (NDSB). The claims specify, among other things, that the NDSB is a quaternary ammonium salt having a nitrogen atom and four groups R1, R2, R3, and R4 – SO^-_3 bound to the nitrogen atom, wherein R1, R2 and R3 can be the same and/or different and are

selected from the group consisting of one or more of methyl, ethyl, propyl, butyl, pentyl, hexyl and derivatives thereof, and R4 is $(CH_2)_n$, wherein n is from 1 to 6. In other words, the NDSB does not have any carbon chains longer than six carbon atoms in length.

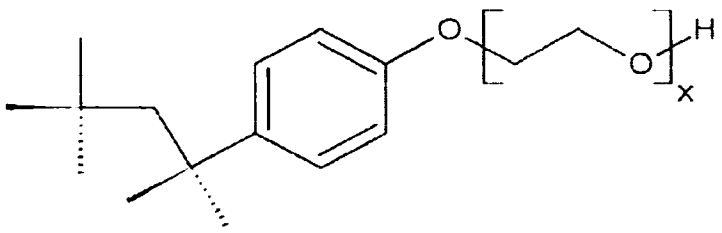
4. We have surprisingly discovered that NDSBs such as those specified in Claims 1, 2, 10, and 12, may be beneficially used as exicipients in final pharmaceutical formulations, particularly in formulations intended for parenteral administration. In these formulations, the NDSBs may function as a stabilizer, a buffering agent, and/or a pH adjusting agent.

5. In contrast to the non-detergent sulfobetaines used in the present invention, the Miyazawa reference discloses sulfobetaines which are said to be "amphoteric surfactants" at Column 3, lines 23 – 32 and Column 4, lines 32 – 42. From this perspective of a person of ordinary skill in the art, these "amphoteric surfactant" sulfobetaines are clearly distinct from the non-detergent sulfobetaines (NDSBs) called for in the present claims.

6. Again, the sulfobetaines in Miyazawa are "amphoteric surfactants." Surfactants (also called "detergents"[1]) are water- soluble amphipathic molecules that possess both a hydrophobic group and a hydrophilic group that allow them to act as excellent solubilization agents. Surfactant molecules contain a hydrophilic polar head group from which extends a long hydrophobic carbon tail. The amphipathic properties of the surfactant molecules allow them to exhibit unique properties in aqueous solutions. The polar (hydrophilic) head groups interact with the hydrogen bonds of the water molecules and the hydrophobic tails aggregate resulting in highly organized spherical structures called micelles.

7. Some typical representative examples of surfactants include:

Octylphenolpoly(ethyleneglycolether)



Molecular Formula: $C_{34}H_{62}O_{11}$ for x =10
Molecular Weight: 647 (for x=10)
Aggregation number: 100-155

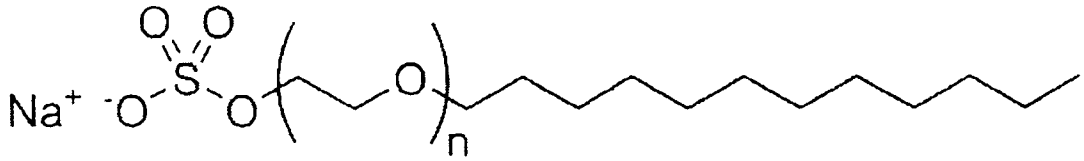
[1] The term "detergent" is more commonly applied in regard to industrial chemicals. The term "surfactant" is more typically applied in the fields of biochemistry and biotechnology.

Average micellar weight: 80,000

Polyoxyethylene (20) cetyl ether

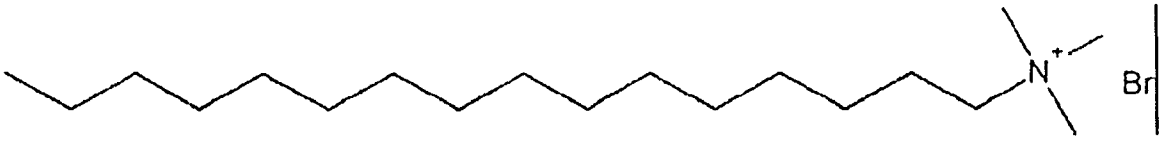
Molecular Formula: $C_{16}H_{33}(OCH_2CH_2)_{20}$-OH
Molecular Weight: 1122
Aggregation number: 70
Average micellar weight: 79,000

Sodium Dodecyl Sulfate



Molecular Formula: $C_{12}H_{25}NaO_4S$
Molecular Weight: 288.38
Aggregation number: 62
Average micellar weight: 18,000

Hexadecyltrimethylammonium bromide



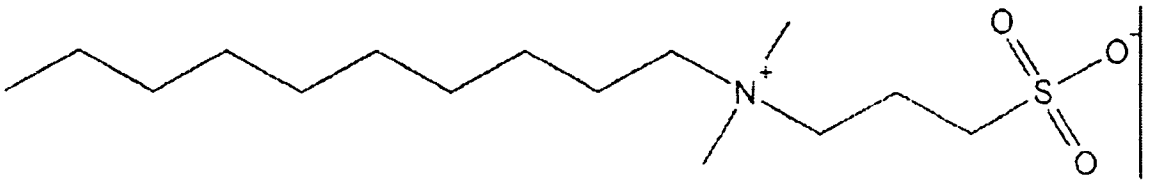
Molecular Formula: $CH_3(CH_2)_{15}N(Br)(CH_3)_3$
Molecular Weight: 364.5
Aggregation number: 61 in H2O; 169 in 13mM KBr
Average micellar weight: 62,000

It may be seen that each of these surfactants has a long hydrophobic carbon tail (substantially longer than six carbon atoms in length) and a correspondingly high molecular weight, from about

300 daltons up to over 1000 daltons. Moreover, individual molecules of these surfactants are observed to aggregate in micelles containing dozens or even hundreds of similar molecules. The overall weight of these aggregated micelles typically ranges from 10,000 up to 100,000 daltons.

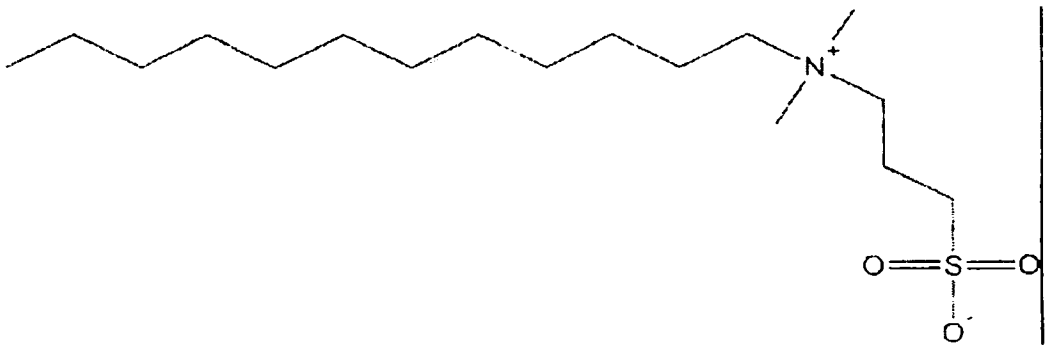
8. More particular examples of surfactant sulfobetaines include:

N-Decyl-N,N-dimethyl-3-ammonio-1-propanesulfonate



Molecular Formula: $CH_3(CH_2)_9N^+(CH_3)_2CH_2CH_2CH_2SO_3$
Molecular Weight: 307.5
Aggregation number: 41
Average micellar weight: 12,600

N-Dodecyl-N,N-dimethyl-3-ammonio-1-propanesulfonate



Molecular Formula: $CH_3(CH_2)_{11}N^+(CH3)_2CH_2CH_2CH_2SO_3^-$
Molecular Weight: 335.5
Aggregation number: 55
Average micellar weight: 18,500

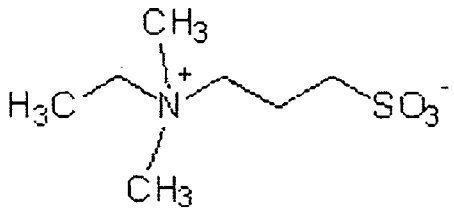
Here again, the molecular weight of these surfactant sulfobetaines are well over 300 daltons and the surfactant sulfobetaines are observed to aggregate in micelles having an overall weight of over 10,000 daltons. Also these surfactant sulfobetaines included long hydrocabon chains well over six carbon atoms in length.

9. While Miyazawa mentions sulfobetaines, the exact length of the carbon chains in these sulfobetaines is not explicitly stated. However, it is notable that the sulfobetaines referred to in Miyazawa are mentioned at Column 3, lines 23 – 32 and Column 4, lines 32 – 42 along side numerous other surfactants having lauryl (12 carbon) substituents, oleyl (18 carbon) substituents, and stearyl (18 carbon) substituents. From the perspective of a person of ordinary skill in the art, Miyazawa's reference to surfactant sulfobetaines in this context would have suggested a large, micelle-forming sulfobetaine molecule having carbon chains of a similiar length.

10. In contrast to these aforementioned surfactant sulfobetaines, the non-detergent sulfobetaines (NDSBs) used according to the present invention are small bipolar compounds containing two oppositely charged polar head groups linked by a short hydrocarbon chain (most commonly a three carbon chain). Due to the short hydrophobic group, such NDSBs cannot aggregate to form micelles. Therefore NDSBs are not considered detergents. In contrast to detergents they are easily removed by dialysis. The NDSBs are a group of zwitterionic compounds that can enhance the recovery of membrane, nuclear and cytoskeleton-associated proteins and aid in refolding proteins found in inclusion bodies and bacterial expression systems. NDSBs have also been used in refolding and renaturation of chemically and thermally denatured proteins.

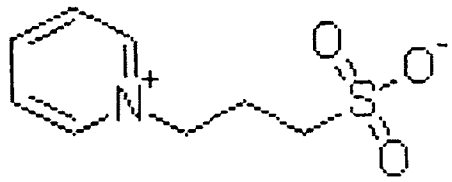
11. Some typical representative examples of the non-detergent sulfobetaines (NDSBs) include:

Dimethylethylammonium propane sulfonate



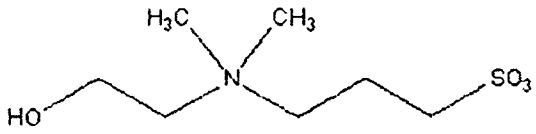
Molecular Weight: 195.3

3-(1-Pyridino)-1-propane sulfonate



Molecular Weight: 201.2

Dimethyl(2-hydroxyethyl)ammonium propane sulfonate



Molecular Weight: 211.3

Again, these compounds do not form micelles in water and therefore cannot be considered to be detergents or surfactants. Such compounds are not disclosed or suggested by the reference to "surfactant sulfobetaines" in the Miyazawa reference.

12. I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true, and further that these statements are made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Dated: October 1, 2010

Vladka Gaberc-Porekar